



PATENT  
Attorney Docket No. 12178/2

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Serial No. : 10/070,480 Confirmation No. 4713  
Applicant(s) : NAKAI et al. Customer No. 23838  
Filed: : Jul 8, 2002  
Art Unit : 1656  
Title: : STEREOSTRUCTURE OF DECARBAMYLASE AND USES  
THEREOF  
Examiner : David J. STEADMAN

**Mail Stop AF**  
Commissioner for Patents  
P.O. Box 1450  
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DECLARATION UNDER RULE 132

I, Takahisa NAKAI, of 7-1-1, Tsutsujigaoka, Tarumi-ku, Kobe-shi, Hyogo, 655-0853, Japan, hereby declare as follows:

I received a degree of a doctor of science from Osaka University in 1994. I have been working in the field of Protein Chemistry and Computational Biology for 19 years. I have been working for KANEKA CORPORATION for 20 years, where I am currently a manager of new product development group in Fine Chemicals Division. I am knowledgeable about techniques for the crystallization of proteins.

I have read and understood the specification of the above-captioned patent application entitled "Stereostructure of Decarbamylase and Uses Thereof." Based on the information contained in the specification, I performed a crystallization of a decarbamylase of SEQ ID NO: 2 using the following steps:

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1. 10  $\mu$ l of a 15 mg/ml solution of decarbamylase (SEQ ID NO: 2) and 10  $\mu$ l of 0.1 M HEPES buffer (pH 7.5) as a precipitant containing 10-15 wt% methoxypolyethylene glycol (PEGMME) 5000 and 0.2 M magnesium chloride were mixed together on a droplet stage.
2. 300  $\mu$ l of the solution having the above-described precipitant composition was sealed as a reservoir solution and crystallization was conducted by a vapor diffusion method at 20-25° C.

Using the aforementioned steps, I successfully obtained crystals of a decarbamylase of SEQ ID NO: 2. PEGMME and magnesium chloride were used instead of PEG and lithium chloride as in Example 1. However, p. 15, lns. 8-12 of the specification describe the use of PEGMME and magnesium chloride as a precipitating agent. By comparing the aforementioned steps and the steps described in Example 1 of the disclosure of the patent application, I conclude that one of ordinary skill in the art would have been able to make crystals of SEQ ID NO: 2 using the disclosure as a guide.

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Dated: May 15, 2006

Signed   
Takahisa NAKAI